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Microbiological Methods in the Ohio District Laboratory

BACTERIOLOGICAL INDICATORS

Coliform group

- Total coliforms are found in the human intestine, in soils, on vegetation, and in industrial wastes. They are used to assess drinking-water or ground-water quality.
- Fecal coliforms are total coliforms that are able to grow at elevated temperatures. They may be of fecal origin.
- *E. coli* is one species in the coliform group, is a natural inhabitant of the gastrointestinal tract of warmblooded animals, and is direct evidence of fecal contamination.

Streptococci

- Fecal streptococci are found in feces; however, some strains are ubiquitous.
- Enterococci is a subset of the fecal streptococci and is a more specific indicator of fecal contamination.

Spore formers

- Certain bacteria produce an environmentally resistant form called a spore.
- *Clostridium perfringens* is present in human and animal wastes:
  - May be used as a surrogate for stress-resistant organisms.
  - May be used as an indicator of fecal contamination from point sources.

Bacterial Methods

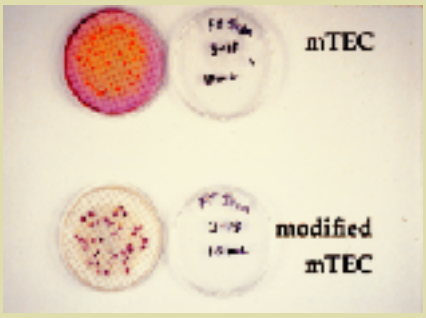

Membrane Filtration–may be done in the field or lab

MEMBRANE FILTRATION

- May be done in the field or lab.
- Uses selective and differential agents in the media.

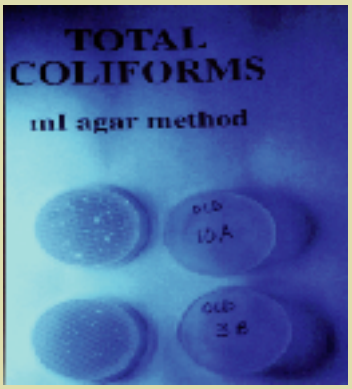
E. COLI mTEC and MODIFIED mTEC

- mTEC – a two step method that detects the breakdown of lactose and includes a test for the enzyme urease (2nd step)– *E. coli* colonies remain yellow.
- Modified mTEC – a one-step method and contains a chromogen for glucuronidase– *E. coli* colonies are magenta.
- Both methods involve two incubation temperatures.



TOTAL COLIFORMS AND *E. COLI*, MI AGAR

- Allows the simultaneous enumeration of *E. coli* and total coliforms and is USEPA approved.
- Contains a fluorogen that reacts with an enzyme found in total coliforms (galactosidase) and a chromogen that reacts with an enzyme found in *E. coli* (glucuronidase).
- Coliform colonies glow under a black light, and *E. coli* colonies appear blue.



CLOSTRIDIUM PERFRINGENS mCP METHOD

- 48-hour holding time before plating.
- Not field friendly-should be done in a laboratory setting by trained personnel.
- *C. perfringens* grown under anaerobic conditions at 42 degrees C.
- Medium contains a chromogen which causes *C. perfringens* colonies to turn bright magenta upon exposure to ammonium hydroxide vapors.


Clostridium perfringens, mCP agar method

ENTEROCOCCI mEI (USEPA METHOD 1600)

- USEPA approved for drinking, recreational, and shelfish-growing waters.
- Requires 24 hour incubation on one medium at 41 degrees C; old method requires 48 hours and two media.
- Chromogen causes enterococci colonies to have blue halos for easy identification.